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<110> KaloBios, Inc.

<120> TRANSACTIVATION SYSTEM FOR MAMMALIAN CELLS

<130> CELA001/01WO

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<170> PatentIn version 3.2

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Pro Thr Leu Ser Pro Val Pro Pro Val Val His Leu Thr Leu Arg Arg 85 90 95

Ala Gly Asp Asp Phe Ser Arg Arg Tyr Arg Arg Asp Phe Ala Glu Met 100 105 110

Ser Ser Gln Leu His Leu Thr Pro Phe Thr Ala Arg Gly Arg Phe Ala 115 120 125

Thr Val Val Glu Glu Leu Phe Arg Asp Gly Val Asn Trp Gly Arg Ile 130 135 140

Val Ala Phe Phe Glu Phe Gly Gly Val Met Cys Val Glu Ser Val Asn 145 150 155 160

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Tyr Leu Asn Arg His Leu His Thr Trp Ile Gln Asp Asn Gly Gly Trp
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Asp Ala Phe Val Glu Leu Tyr Gly Pro Ser Val Arg Pro Leu Phe Asp 195 200 205

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Arg Arg Asp Phe Ala Glu Met Ser Ser Gln Leu His Leu Thr Pro Phe 65 70 75 80

Thr Ala Arg Gly Arg Phe Ala Thr Val Val Glu Glu Leu Phe Arg Asp 85 90 95

Gly Val Asn Trp Gly Arg Ile Val Ala Phe Phe Glu Phe Gly Gly Val

Met Cys Val Glu Ser Val Asn Arg Glu Met Ser Pro Leu Val Asp Asn 115 120 125

Ile Ala Leu Trp Met Thr Glu Tyr Leu Asn Arg His Leu His Thr Trp

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Phe Glu Glu Thr Glu Glu Pro Asp Phe Thr Ala Leu Cys Gln Lys Leu 50 60

Lys Ile Pro Asp His Val Arg Glu Arg Ala Trp Leu Thr Trp Glu Lys 65 70 75 80

Val Ser Ser Val Asp Gly Val Leu Gly Gly Tyr Ile Gln Lys Lys 85 90 95

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Gln Val His Gly Val Ile Gln Ala Ala Gln Pro Ser Val Ile Gln Ser 65 70 75 80

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Ser Gln Glu Ser Val Asp Ser Val Thr Asp Ser Gln Lys Arg Arg Ile 100 105 110

Leu Ser Arg Arg Pro Ser Phe Arg Lys Ile Leu Asn Asp Leu Ser Ser 115 120 125

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<212> DNA

<213> Artificial

<220>

<223> primer

<400> 22

cccgaattcg ccgccaccat gaccatggac tctggagcag aca 43

<210> 23

<211> 30

<212> DNA

<213> Artificial

<220>

<223> primer

<400> 23

gtcgacccaa attaatctga tttgtggcag

30

Primer 14: GTCAAGCAAGCTTGCCGCCACCATGAGACATATTATCTGCCACGG (SEQ ID NO: 24)

Primer 15: CGCAGTCTCGAGTTATGGCCTGGGGCGTTTACAGCTC (SEQ ID NO: 25)

Primer 16: CACCTACCCTTCACGAACTGCATGATTTAGACGTGACGGCC (SEQ ID NO: 26)

Primer 17: GGCCGTCACGTCTAAATCATGCAGTTCGTGAAGGGTAGGTG (SEQ ID NO: 27)

Primer 18: CGGAGGTGATCGTTACCGGCCACGAGGCTGGCTTTCCAC (SEQ

ID NO: 28)

Primer 19: GTGGAAAGCCAGCCTCGTGGCCGGTAAGATCGATCACCTCCG (SEQ

ID NO: 29)

Primer 20: GTCAAGCAAGCTTGCCGCCACCATGACCATGGAATCTGGAGC (SEQ

ID NO: 30)

Primer 21: CGCAGTGGATCCTTAATCTGATTTGTGGCAGTAAAGG (SEQ ID NO: 31)

Primer 22: GTCATTCAAAATTTTCCTGAAGGAAGGCCTCCTTGAAAG (SEQ ID

NO: 32)

Primer 23: TCTTTCAAGGAGGCCTTCCTTCAGGAAAATTTTGAATGAC (SEQ ID

NO: 33)

Primer 24: GGCATTCCAAGCTTACTGTTGGTAAAGCCGCCACCATGGAGGCTTGGG

AGTGTTTGG (SEQ ID NO: 34)

Primer 25: GATCGACTCTAGATCATTCCCGAGGGTCCAGGCCGG (SEQ ID NO:

35)

Primer 26: TAAAGCCACCATGGCTCAAGCTGGGAGAACAGGGTATG (SEQ ID

NO: 36)

Primer 27: GATCGACTCTAGATCACTTGTGGCCCAGGTAGGTACCC (SEQ ID

NO: 37)

Primer 28: GTGGGAGATGTGGACGCCGCGGCCGCGCGCGCGCGCCGTGCCAC

CTGTGGTCC (SEQ ID NO: 38)

Primer 29: GGACCACAGGTGGCACGGGGCTCGCGGCCGCGCGCGCGTCCAC

ATCTCCCAC (SEQ ID NO: 39)

(From Figure 10) Nucleotide coding sequence of Ela cDNA (SEQ ID NO: 40):

1 aagettgeeg eeaceatgag acatattate tgeeaeggag gtgttattae HindIII M R H I I C H G G V I

51 cgaagaaatg gccgccagtc ttttggacca gctgatcgaa gaggtactgg T E E M A A S L L D Q L I E E V L

101 ctgataatct tccacctcct agccattttg aaccacctac ccttcacgaa A D N L P P P S H F E P P T L H E

151 ctgtatgatt tagacgtgac ggcccccgaa gatcccaacg aggaggcggt L \underline{Y} D L D V T A P E D P N E E A

201 ttegcagatt ttteccgact etgtaatgtt ggeggtgeag gaagggattg V S Q I F P D S V M L A V Q E G I

251 acttactcac ttttccgccg gcgcccggtt ctccggagcc gcctcacctt D L L T F P P A P G S P E P P H L

- 301 tcccggcagc ccgagcagcc ggagcagaga gccttgggtc cggtttctat S R Q P E Q P E Q R A L G P V S
- 351 gccaaacctt gtaccggagg tgatcgatct tacctgccac gaggctggct M P N L V P E V I D L T C H E A G
- 401 ttccacccag tgacgacgag gatgaagagg gtgaggagtt tgtgttagat FPPSDDEDEEGEFVLD
- 451 tatgtggagc accccgggca cggttgcagg tcttgtcatt atcaccggag Y V E H P G H G C R S C H Y H R
- 501 gaatacgggg gacccagata ttatgtgttc gctttgctat atgaggacct R N T G D P D I M C S L C Y M R T
- 551 gtggcatgtt tgtetacagt cetgtgtetg aacetgagee tgageeegag C G M F V Y S P V S E P E P E P E
- 601 ccagaaccgg agcetgeaag acctacccge cgtcctaaaa tggcgcctgc P E P A R P T R R P K M A P
- 651 tatcctgaga cgcccgacat cacctgtgtc tagagaatgc aatagtagta A I L R R P T S P V S R E C N S S
- 701 cggatagetg tgaeteeggt cettetaaca caceteetga gatacaceeg T D S C D S G P S N T P P E I H P
- 751 gtggtcccgc tgtgccccat taaaccagtt gccgtgagag ttggtgggcg V V P L C P I K P V A V R V G G
- 801 tegecagget gtggaatgta tegaggaett gettaaegag eetgggeaae R R Q A V E C I E D L L N E P G Q
- 851 ctttggactt gagctgtaaa cgccccaggc cataactcga g P L D L S C K R P R P - Xhol

(From Figure 11) Nucleotide coding sequence of ElA mutant Y47H (SEQ ID NO: 41):

- 1 <u>aagettgeeg</u> ceaceatgag acatattate tgeeaeggag gtgttattae *HindIII* M R H I I C H G G V I
- 51 cgaagaaatg geegeeagte t
tttggacea getgategaa gaggtaetgg ${\tt T}$ E E M A A S L L D Q L I E E V L
- 101 ctgataatct tccacctcct agccattttg aaccacctac ccttcacgaa A D N L P P P S H F E P P T L H E
- 151 ctgcatgatt tagacgtgac ggccccgaa gatcccaacg aggaggcggt L H D L D V T A P E D P N E E A
- 201 ttegcagatt tttecegaet etgtaatgtt ggeggtgeag gaagggattg V S Q I F P D S V M L A V Q E G I
- 251 acttactcac ttttccgccg gcgcccggtt ctccggagcc gcctcacctt D L L T F P P A P G S P E P P H L
- 301 tcccggcagc ccgagcagcc ggagcagaga gccttgggtc cggtttctat S R Q P E Q P E Q R A L G P V S
- 351 gccaaacctt gtaccggagg tgatcgatct tacctgccac gaggctggct

MPNL VPE VID LTCH EAG

- 401 ttccacccag tgacgacgag gatgaagagg gtgaggagtt tgtgttagat FPPSDDEDEEGEFVLD
- 451 tatgtggagc accccgggca cggttgcagg tcttgtcatt atcaccggag Y V E H P G H G C R S C H Y H R
- 501 gaatacgggg gacccagata ttatgtgttc gctttgctat atgaggacct R N T G D P D I M C S L C Y M R T
- 551 gtggcatgtt tgtctacagt cctgtgtctg aacctgagcc tgagcccgag C G M F V Y S P V S E P E P E P E
- 601 ccagaaccgg agcctgcaag acctaccgc cgtcctaaaa tggcgcctgc P E P A R P T R R P K M A P
- 651 tatcctgaga cgcccgacat cacctgtgtc tagagaatgc aatagtagta A I L R R P T S P V S R E C N S S
- 701 eggatagetg tgaeteeggt cettetaaca caceteetga gatacaceeg T D S C D S G P S N T P P E I H P
- 751 gtggtcccgc tgtgccccat taaaccagtt gccgtgagag ttggtgggcg V V P L C P I K P V A V R V G G
- 801 tcgccaggct gtggaatgta tcgaggactt gcttaacgag cctgggcaac R R Q A V E C I E D L L N E P G Q
- 851 ctttggactt gagctgtaaa cgccccaggc cataactcga g P L D L S C K R P R P - *XhoI*

(From Figure 12) Nucleotide coding sequence of hamster CREB-B cDNA (SEQ ID NO: 42):

- 51 tggagatgct gctgtaacag aagctgaaaa tcaacaaatg acagctcaag S G D A A V T E A E N Q Q M T A Q
- 101 cccaaccaca gattgccaca ttagcccagg tatccatgcc agcagctcat A Q P Q I A T L A Q V S M P A A H
- 151 gegacateat etgeteceae tgtaacetta gtgeagetge ecaatgggea ATSSAPTVTLVQLPNG
- 201 gacagtccaa gtccatggag ttattcaggc ggcccagcca tcagttattc Q T V Q V H G V I Q A A Q P S V I
- 251 agtetecaca agtecaaaca gtteagtett eetgtaagga ettaaaaaga Q S P Q V Q T V Q S S C K D L K R
- 301 cttttctccg gaactcagat ttcaactatt gcagaaagtg aggattcaca L F S G T Q I S T I A E S E D S
- 351 ggaatctgtg gatagtgtaa ctgattccca aaagcgaagg gaaattcttt Q E S V D S V T D S Q K R R E I L
- 401 caaggaggcc ttcctacagg aaaattttga atgacttatc ttctgatgca

S R R P S Y R K I L N D L S S D A

- 451 ccaggggtgc caaggattga agaagaaaag tcggaagagg agacttcagc P G V P R I E E E K S E E E T S
- 501 ccctgccatc accactgtga cagtgccaac tccgatttac cagacaagca A P A I T T V T V P T P I Y Q T S
- 551 gtgggcagta tattgccatt acccagggag gagctataca gctggctaac S G Q Y I A I T Q G G A I Q L A N
- 601 aatggtaccg atggggtaca gggccttcag acattaacca tgaccaatgc N G T D G V Q G L Q T L T M T N
- 651 agetgecaet cageegggta ceaetattet acagtatgea cagaceaetg A A A T Q P G T T I L Q Y A Q T T
- 701 atggacagca gattetagtg cccagcaacc aagttgttgt tcaagctgcc D G Q Q I L V P S N Q V V V Q A A
- 751 tetggegatg tacaaacata ccaaattegt acageaceca etageaceat S G D V Q T Y Q I R T A P T S T
- 801 cgcccctgga gttgttatgg catcctcccc agcacttcct acgcagcctg I A P G V V M A S S P A L P T Q P
- 851 ctgaagaagc agcccggaag agagaggttc gtctaatgaa gaacagggaa A E E A A R K R E V R L M K N R E
- 901 gcagcaagag aatgtcgtag aaagaagaaa gaatatgtga aatgtttaga AARECRRKKEYVKCL
- 951 gaacagagtg gcagtgettg aaaaccaaaa caagacattg attgaggagc E N R V A V L E N Q N K T L I E E
- 1001 taaaagcact taaggacctt tactgccaca aatcagatta aggatcc L K A L K D L Y C H K S D BamHI

(From Figure 13) Nucleotide coding sequence of hamster CREB-B mutant Y134F (SEQ ID NO: 43):

- 51 tggagatget getgtaacag aagetgaaaa teaacaaatg acageteaag S G D A A V T E A E N Q Q M T A Q
- 101 cccaaccaca gattgccaca ttagcccagg tatccatgcc agcagctcat A Q P Q I A T L A Q V S M P A A H
- 151 gcgacatcat ctgctcccac tgtaacctta gtgcagctgc ccaatgggca A T S S A P T V T L V Q L P N G
- 201 gacagtecaa gtecatggag ttatteagge ggeecageea teagttatte Q T V Q V H G V I Q A A Q P S V I
- 251 agtetecaea agteeaaaca gtteagtett eetgtaagga ettaaaaaga $Q\ S\ P\ Q\ V\ Q\ T\ V\ Q\ S\ S\ C\ K\ D\ L\ K\ R$
- 301 cttttctccg gaactcagat ttcaactatt gcagaaagtg aggattcaca L F S G T Q I S T I A E S E D S

- 351 ggaatctgtg gatagtgtaa ctgattccca aaagcgaagg gaaattcttt Q E S V D S V T D S Q K R R E I L
- 401 caaggaggee tteeteeagg aaaattttga atgaettate ttetgatgea S R R P S F R K I L N D L S S D A
- 451 ccaggggtgc caaggattga agaagaaaag tcggaagagg agacttcagc P G V P R I E E E K S E E E T S
- 501 ccctgccatc accactgtga cagtgccaac tccgatttac cagacaagca A P A I T T V T V P T P I Y Q T S
- 551 gtgggcagta tattgccatt acccagggag gagctataca gctggctaac S G Q Y I A I T Q G G A I Q L A N
- 601 aatggtaccg atggggtaca gggccttcag acattaacca tgaccaatgc N G T D G V Q G L Q T L T M T N
- 651 agetgecaet cageegggta ceaetattet acagtatgea cagaceaetg AAATQPGTTILQYAQTT
- 701 atggacagca gattctagtg cccagcaacc aagttgttgt tcaagctgcc D G Q Q I L V P S N Q V V V Q A A
- 751 tetggegatg tacaaacata ccaaattegt acageaceca etageaceat S G D V Q T Y Q I R T A P T S T
- 801 cgcccctgga gttgttatgg catcctcccc agcacttcct acgcagcctg I A P G V V M A S S P A L P T Q P
- 851 ctgaagaagc agcccggaag agagaggttc gtctaatgaa gaacagggaa A E E A A R K R E V R L M K N R E
- 901 gcagcaagag aatgtcgtag aaagaagaaa gaatatgtga aatgtttaga AARECRRKKEYVKCL
- 951 gaacagagtg gcagtgcttg aaaaccaaaa caagacattg attgaggagc ENRVAVLENQNKTLIEE
- 1001 taaaageact taaggacctt tactgccaca aatcagatta aggatcc L K A L K D L Y C H K S D BamHI

(From Figure 14) Nucleotide coding sequence of Elb-19K (SEQ ID NO: 44):

- 1 <u>aagett</u>actg ttggtaaage egecaceatg gaggettggg agtgtttgga HindIII M E A W E C L
- 51 agatttttct gctgtgcgta acttgctgga acagagctct aacagtacct E D F S A V R N L L E Q S S N S T
- 101 cttggttttg gaggtttctg tggggctcat cccaggcaaa gttagtctgc S W F W R F L W G S S Q A K L V C
- 151 agaattaagg aggattacaa gtgggaattt gaagagcttt tgaaatcctg RIKEDYKWEFEELLKS

- 201 tggtgagetg tttgattett tgaatetggg teaceaggeg etttteeaag CGELFDSLNLGHQALFQ
- 251 agaaggtcat caagactttg gatttttcca caccggggcg cgctgcggct E K V I K T L D F S T P G R A A A
- 301 gctgttgctt ttttgagttt tataaaggat aaatggagcg aagaaaccca A V A F L S F I K D K W S E E T
- 351 tetgageggg gggtacetge tggattttet ggccatgcat etgtggagag H L S G G Y L L D F L A M H L W R
- 401 cggttgtgag acacaagaat cgcctgctac tgttgtcttc cgtccgcccg A V V R H K N R L L L S S V R P
- 451 gcgataatac cgacggagga gcagcagcag cagcaggagg aagccaggcg A I I P T E E Q Q Q Q E E A R
- 501 geggeggeag gageagagee catggaacee gagageegge etggaeeete R R R Q E Q S P W N P R A G L D P
- 551 gggaatga<u>tc taga</u> R E - *XbaI*

(From Figure 15) Nucleotide coding sequence of hamster Bcl2 deletion mutant (SEQ ID NO: 45):

- NCOI
 1 ccatggctca agctgggaga acagggtatg ataaccgaga gatcgtgatg
 M A Q A G R T G Y D N R E I V M
- 51 aagtacatcc attataagct gtcacagagg ggctacgagt gggatgtggg K Y I H Y K L S Q R G Y E W D V
- 101 agatgtggac gccgcggccg cggccgcgag ccccgtgcca cctgtggtcc G D V D A A A A A A S P V P P V V
- 151 acctgaccct ccgccgggct ggggatgact tctcccgtcg ctaccgtcgc H L T L R R A G D D F S R R Y R R
- 201 gacttcgcgg agatgtccag tcagctgcac ctgacgccct tcaccgcgag D F A E M S S Q L H L T P F T A
- 251 gggacgettt getaeggtgg tggaggaact etteagggat ggggtgaact R G R F A T V V E E L F R D G V N
- 301 gggggaggat tgtggccttc tttgagttcg gtggggtcat gtgtgtggag W G R I V A F F E F G G V M C V E
- 351 agegteaaca gggagatgte acceetggtg gacaacateg ceetgtggat

S V N R E M S P L V D N I A L W

- 401 gaccgagtac ctgaaccggc atctgcacac ctggatccag gataacggag M T E Y L N R H L H T W I Q D N G
- 451 getgggaege atttgtggaa etgtaeggee eeagtgtgag geetetgttt G W D A F V E L Y G P S V R P L F
- 501 gatttetett ggetgtetet gaagaeeetg eteageetgg eeetggtegg DFSWLSLKTLLSLALV
- 551 ggcctgcatc actetgggta cctacctggg ccacaagtga tctaga G A C I T L G T Y L G H K XbaI

Primer 30: CGCAGTACTAGTTTATGGCCTGGGGCGTTTACAGCTC (SEQ ID NO:46)

Primer 31: GAGCTATTCCAGAAGTAGTG (SEQ ID NO:47)